EXOSOME DEPENDENT TRAFFICKING OF STRESS-INDUCIBLE PROTEIN 1 IN ASSOCIATION WITH CELLULAR PRION PROTEIN

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The physiological functions of PrP^c are under intensive investigation, particularly those associated to brain development. We have described that PrP^c binds Stress Inducible Protein 1 (STI1) inducing neuritogenesis via MAPK. Furthermore, STI1 can be secreted by astrocytes, even though it lacks a secretory signal sequence. Since it has been previously shown that PrP^c can be secreted by exosomes, we decided to examine whether this secretory pathway contributes to STI1 release. Exosomes were purified from conditioned medium from wild-type (*Prnp*$^{+/+}$) and PrP^c ablated (*Prnp*$^{0/0}$) primary astrocytes. Quantification of exosomal release was evaluate by western blot, with positive marks for transferrin receptor, HSP70, HSP90 and PrP^c, and by measuring exosome associated acetyl-cholinesterase activity. Conversely, STI1 secretion is independent on the classical secretory pathway mediated by the Golgi apparatus, since astrocyte treatment with Brefeldin A and Monensin had no blocking effect. GFP-STI1 is also secreted by exosomes and we could further show that cultured hippocampal neurons are able to bind and internalize it. Additionally, we show that STI1 released in exosomes activates the MAPK signaling pathway in *Prnp*$^{+/+}$ hippocampal neurons but not in *Prnp*$^{0/0}$ ones. We also demonstrate that the protein content of *Prnp*$^{0/0}$ exosomes is significantly lower than *Prnp*$^{+/+}$ and is not capable of inducing MAPK activation. Taken together these findings indicate that PrP^c expression in astrocytes can modulate the secretion of exosomes or their protein content. Moreover, exosomes containing STI1 are able to modulate a signaling pathway previously associated to neuronal differentiation. Supported by Fapesp and HHMI.